

**Claims**

1. A nucleotide sequence encoding a WT1 antisense regulatory region comprising at least a portion of the sequence shown in SEQ.1 or at least a portion of a variant, due to base substitutions, deletions, and/or additions, of the sequence shown in SEQ.1.
2. A nucleotide sequence according to claim 1 which encodes a WT1 antisense regulatory region negative regulatory element (NRE).
3. A WT1 antisense regulatory region negative regulatory element (NRE) comprising at least a portion of the nucleotide sequence shown in SEQ.2 or at least a portion of a variant, due to base substitutions, deletions, and/or additions, of the sequence shown in SEQ.2.
4. A WT1 antisense regulatory region NRE according to claim 3 wherein the NRE comprises the sequence shown in bold in SEQ. 2, or variants of such a sequence due to base substitutions, deletions and/or additions.
5. A nucleotide sequence or NRE according to any preceding claim wherein the nucleotide sequence is a DNA sequence.
6. An RNA sequence encoded by a nucleotide sequence according to any preceding claim.
7. A method of disease diagnosis and prognosis in a subject diagnosed with a Wilms' tumour cancer, the method comprising determining the differentially methylated state of a specific nucleotide sequence or sequences in the subject, or in a sample derived from the subject.
8. A method according to claim 7 wherein the specific nucleotide sequence or sequences form part of the WT1 antisense regulatory region (ARR).

9. A method according to claim 7 or claim 8, comprising determining the methylation state of a negative regulatory element (NRE) or an ARR of a WT1 gene in a sample isolated from the subject, and correlating the methylation state of the NRE or ARR with the diagnosis and expected long-term recovery prognosis of the subject.
10. A method according to claim 9 wherein hypermethylation of the NRE or ARR indicates that the subject has a positive long term recovery prognosis, and hypomethylation of the NRE or ARR indicating that the subject is predisposed to relapsing after treatment.
11. A method according to claim 7 or 8, wherein hypomethylation of the specific nucleotide sequence or sequences indicates that the subject has a positive long term recovery prognosis, and hypermethylation of the specific nucleotide sequence or sequences indicates that the subject is predisposed to relapsing after treatment.
12. A method according to any one claims 7 to 11 wherein the NRE is a nucleotide sequence according to any one of claims 1 to 6.
13. A method according to any one of claims 7 to 12 wherein the methylation state is detected by restriction digest analysis.
14. A method according to claim 13 wherein at least enzyme Bsh1236I is used to restrict the NRE.
15. A method according to any one of claims 7 to 12 wherein the methylation state is detected using a PCR-based assay system.
16. A method according to claim 15 wherein the PCR assay system uses at least one of the following primers to amplify a region of nucleotide sequence:  
  
Tf: 5'-GGGTGGAGAAGAAGGATATATTTAT-3'.

Tr: 5'-TAAATATCAAATTAATTTCTCATCC-3'.

TtN: 5'-GATATATTTATTTATTAGTTTTGGT-3' (nested primer).

TtN: 5'-AAACCCCTATAATTTACCCTCTTC-3' (nested primer).

17. A method according to claim 16 wherein the amplified nucleotide sequence is cloned and sequenced.
18. A probe comprising a nucleotide sequence according to any one of claims 1 to 6.
19. A diagnostic kit, assay, or monitoring method using a nucleotide sequence according to any one of claims 1 to 6 or a probe according to claim 18.
20. A diagnostic kit, assay, or monitoring method using a method according to any one of claims 7 to 17.
21. A method of cancer detection in a subject or in a sample isolated from the subject comprising detection of the methylation state of a specific nucleotide sequence or sequences.
22. A method according to claim 21 comprising correlating the methylation state of the specific nucleotide sequence or sequences with the presence or absence of cancer cells in the subject.
23. A method according to claim 22 wherein hypomethylation of the specific nucleotide sequence or sequences indicates the presence of cancer cells in the subject.
24. A method of cancer detection in cells derived from a subject comprising detection of tumour-specific alteration of genomic imprinting.
25. A method according to claim 24 comprising the detection of tumour-specific relaxation of genomic imprinting by determining the methylation state of a specific nucleotide sequence.
26. A method according to claim 24 or claim 25 wherein the tumour-specific alteration of genomic imprinting is detected by reverse transcription-PCR (RT-PCR).

27. A method according to any one of claims 24 to 26 wherein the cancer is Wilms' Tumour (WT).
28. A method according to claim 27 comprising detection of the relaxation of genomic imprinting of the antisense WT-1 RNA sequence.
29. A method according to claim 28 wherein the RT-PCR uses two primers, designed to anneal to the tumour-specific gene sequence on opposite sides of an allelic polymorphism which introduces a restriction site in one allele only.
30. A method according to claim 29 wherein the RT-PCR uses the following primer pair
- Primer 1: WT18 CTTAGCACTTTCTTCTTGGC
- Primer 2: WITKBF2 TTGCTCAGTGATTGACCAGG.
31. A method of treating a subject with a specific cancer comprising altering the genomic imprinting of a tumour-specific gene.
32. A method according to claim 31 wherein the genomic imprinting of a tumour-specific gene is altered by altering the methylation state of a specific nucleotide sequence.
33. A method according to claim 31 or claim 32 wherein the genomic imprinting is altered to relax the genomic imprinting of the tumour-specific gene.
34. A method according to claim 31 or claim 32 wherein the genomic imprinting is altered to reverse the relaxation of the genomic imprinting of the tumour-specific gene.
35. A diagnostic kit, assay or a monitoring method using a method according to any one of claims 24 to 30.
36. A method of detection of the methylation state of a WT1 antisense regulatory region comprising detection of a tumour-specific alteration of genomic imprinting using a method according to any one of claims 21 to 30 and correlating a detected alteration in relaxed genomic imprinting with differential methylation of the WT1 antisense regulatory region.

- 37. A method according to claim 36 wherein the alteration in genomic imprinting is a relaxation in genomic imprinting.**
- 38. Method of treatment comprising selecting a particular course of therapy on the basis of the results of a method according to any preceding claim.**